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Spontaneous Neoplasms in Fishes. I. Osteochondroma in the Jewelfish, *Hemichromis bimaculatus*.[1]

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Plates I-V; Text-figures 1 & 2.

INTRODUCTION.

Only a few chondromas and related tumors have been reported, all in European-bred fishes. Fiebiger (1909) described a massive osteochondroma on a carp (*Cyprinus*) which grew into the cranial cavity, causing the brain to be compressed and laminated. Although no functional disturbances were noted in the live fish, Fiebiger reported that the olfactory lobes, certain cranial nerves and hypophysis were missing. A chondroma, also in a carp, was reported and figured (credited to Muslow) by Plehn (1924). A longitudinal section of the head showed the tumor extending ventrally over the roof of the mouth and posteriorly against the brain, causing it to be deflected upward and at right angles to the brain stem. Thomas (1931), in his excellent review on fish tumors, recorded two cases described by Surbeck in 1917 and 1921. The first was an enchondroma which developed in a barbel (*Barbus*) as a bilobed structure between the maxilla and the right operculum. The second was a chondrofibroma which was attached to the belly of a pike. This growth was a massive, pediculated, saccular tumor larger than two fists and weighing 800 grams.

No detailed histological or cytological descriptions were given of the tumors reported above. Neither was there any information given as to the possible cause or causes of the growths. The present contribution is concerned with gross and histological descriptions of an osteochondroma on the operculum and adjacent structures of an aquarium-bred African jewelfish, *Hemichromis bimaculatus*. This fish was found among about 100 normal jewelfish in a 150 gallon aquarium in the Department of Animal Behavior of the American Museum of Natural History. We are indebted to Dr. Lester R. Aronson for this specimen.

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GROSS DESCRIPTION OF THE TUMOR.

When the tumor was first observed it appeared as a small swelling at the anterior edge of the right operculum. The growth was comparatively rapid, for within two months it attained the large size shown in Text-figure 1 and Plate I, Figure 1. The fish was a male and measured 52 mm. in standard length; the tumor mass measured 13 mm. long, 6 mm. at its widest part and from 3 to 4 mm. high. When examined with the low power binocular, the surface of the growth was more or less smooth and richly supplied with branches of the right first afferent artery (Text-figure 1). The structures involved were the opercle, subopercle, peropercle, interopercle, brachiostegals, inferior parts of the maxillary and cheek bones. The anterior-most gill-raker of the first gill arch also had a smaller but separate cartilaginous nodule, which measured about 3 mm. in diameter. It was uncertain whether this separate growth arose *de novo* or whether some infiltration had taken place, either directly or through the blood stream.

HISTOLOGICAL DESCRIPTION OF THE TUMOR.

The tumor, together with the adjacent structures, was extirpated and pieces were fixed in Zenker's. They were embedded in paraffin, sectioned at four microns and stained with iron-hematoxylin with and without eosin, Delafield's hematoxylin with eosin, Mallory's triple stain, Giemsa's stain, and with methylene blue.

The tumor was an osteochondroma, the major part of which consisted of hyalin cartilage arranged in a more or less irregular pattern. (Plates I-V). The typical arrangement of cartilage cells with opposed surfaces was not evident. Some tranformation into osteoid tissue had taken place (Plate II, Figure 6). The tissue was well supplied with nourishment from the numerous blood vessels which accompanied its fibrous stroma (Plate IV, Figure 9). The

TEXT-FIGURE 1. Sketch of the ventral-lateral aspect of the head of the jewelfish showing the blood vessels on the surface of the osteochondroma. The main branch emerging from the branchial region is derived from the right first afferent artery. Enlarged about 4 ×.

intercellular substance was variable, being very dense in some areas and light in others (Plate I, Figure 3). In regions where the cartilage cells appeared to be dividing rapidly, there was little intercellular material and the isogenic groups of tumor cells were closer (Plate II, Figures 5 and 6). Both interstitial and appositional growth took place to form the tumor.

The epithelium overlying the growth was still intact, slightly thickened in some areas, but otherwise normal in appearance (Plate I, Figure 2). The corial layer was edematous and contained only a few melanophores.

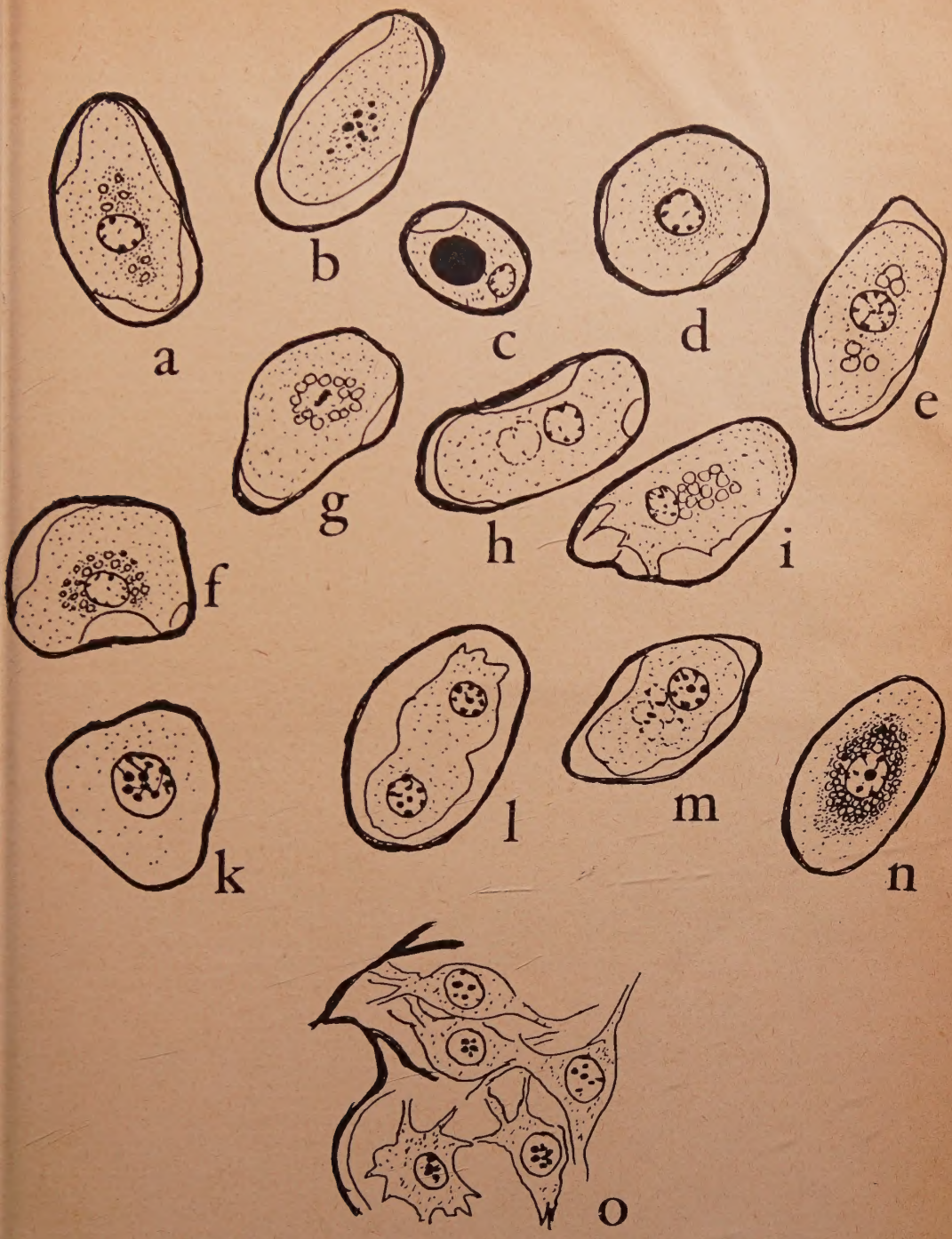
The thickened periosteum was basophilic (Plate I, Figure 2) while the subperiosteum was mainly acidophilic (Plate II, Figure 4). Numerous large and small, thin and thick walled blood vessels were present in the subperiosteal layer. The vessels, however, were never completely engorged and often contained granular debris. The transformation of the cellular elements of the subperiosteum into typical chondrocytes at the periphery of the tumor was evident. Such cells were responsible for the appositional growth. In some regions the gradual transition from collagenous fibers, made up of spindle-shaped cells (fibroblasts), into cartilage was visible (Plate IV, Figure 10). In other and more highly vascularized areas the various cellular stages in bone formation were found. The elements involved in the process were stellate-shaped (osteoblasts) which passed from the subperiosteal layer into the tumor mass. These cells (Text-figure 2, n; Plate V, Figures 11 and 12), interconnected by their dendritic processes, formed a network. They laid down a hyalin-like ground substance, which, however, was not calcified. (Such tissue is referred to as osteoid. In fishes osteoid tissue enters into the composition of scales and in other structures of the head, including the operculum).

The fully developed cartilage cells (Plate II, Figures 5 and 6) were irregular in shape, size and in their mass arrangement. In some areas the cells were large with clearly defined capsules and had a great deal of intercellular substance between them. In other areas, particularly at the periphery of the tumor, the cells were smaller and isogenic groups were more clearly separated from each other (Plate II, Figure 6). There was marked evidence of cellular activity (interstitial tissue growth), although no mitotic figures were noted. Many cells were binucleated (Text-figure 2, k) and in some cases two or even three cells were present within the same territorial matrix. Although the majority of cells were spherical many variations were found. Those toward the periphery of the tumor were more or less flattened in a plane parallel with the surface. On the borderline of the cartilage and periosteum there were intermediate forms between the cartilage cells and the fibroblasts (Plate III, Figure 8). The body of the cartilage cells, in the fixed and stained sections, did not always fill the cartilage cavity which it occupied in the interstitial substance. A few cells had dendritic processes (Text-figure 2, i; Plate IV, Figure 9) but whether or not these processes extended into the interstitial substance was not determined, although the indication was that they did.

The nuclei of the mature chondrocytes were comparatively large and usually vesicular (Text-figure 2; Plates I-V). The cytoplasm was granular and in the region surrounding the nucleus was denser and often contained minute vacuole-like structures which in some cells were small and numerous and in others larger and few (Plate II, Figure 5; Text-figure 2). These vacuoles were present in all preparations, regardless of fixative or stain used. It is believed that the presence of these vacuoles indicates the secretory activity of the cell, the contents of the vacuole passing into the interstitial area. Other cells contained comparatively large concretions (chondrin?) (Text-figure 2, c) which stained similar to the intercellular material.

DISCUSSION.

The osteochondroma of the jewelfish is strikingly similar histologically to comparable growths in man (see Ewing, 1940). The cause of the tumor on the fish was not determined. It was the only abnormal specimen in about a hundred of the same species



TEXT-FIGURE 2. Figures **a-i** inclusive are chondrocytes stained with Giemsa's stain: **a**, **e** and **f-i** show different manifestations of the nucleus together with the vacuole-like structures around it; **b**, cell with basophilic staining granules which may be chromatin; **c**, cell with concretion (chondrin?) that stains like the intercellular substance. Figures **k-n** inclusive were stained with iron-hematoxylin: **k**, a typical chondrocyte; **l**, binucleate cell within the same capsule; **m**, cell with five vacuole-like structures in the region of the nucleus; **n**, cell with dense cytoplasm, containing many minute vacuole-like structures, surrounding the nucleus. Figure **o**, a group of osteoblast cells. Note the beginning of bone formation. About 800 ×.

and strain that were kept in a 150 gallon aquarium at the Department of Animal Behavior of the American Museum of Natural History. In all other respects the fish appeared to be in good health; the gills and internal organs showed no obvious disturbances. There was no evidence that heredity, hormonal disturbance, nutritional deficiency, virus, bacteria or other plant and animal parasites were involved as possible causative agents of this osteochondroma. However, the tumor may have resulted from traumatic response following an injury to the fish's operculum earlier in life. Periosteum is readily capable of producing cartilage whenever new formation of this tissue is required, and cartilaginous tumors might possibly arise from cells which produce bone by way of cartilage.

Summary.

A spontaneous osteochondroma on the right operculum and associated structures of a jewelfish, *Hemichromis bimaculatus*, was composed of irregularly arranged hyalin cartilage and osteoid tissue. It was well supplied with blood vessels. Both interstitial and appositional cell growth were responsible for the increase in size of the tumor. The transformation of the cellular elements of the connective tissue in the subperiosteal region into chondrocytes at the periphery of the tumor was clearly evident. In several regions stellate-shaped cells (osteoblasts) passed into the mass of the tumor from the subperiosteal layer and formed a network around which hyalin-like osteoid material was deposited. The major part of the tumor consisted of hyalin cartilage. Cytological details of the chondrocytes were given. The causative agent of the osteochondroma was not determined but it may have resulted from a fractured operculum in early life.

References.

Ewing, James.

1940. Neoplastic Diseases. 4th Edition; 1160 pp., 581 illustrations. Philadelphia and London. W. B. Saunders Co.

Fiebiger, J.

1909. Ueber Hautgeschwülste bei Fischen, nebst Bemerkungen über die Pokenkrankheiten der Karpfen. *Zeitschrf. f. Krebsforsch.* 7: 165.

Plehn, M.

1924. Prakticum der Fischkrankheiten. 179 pp., 21 pls., 173 text-figures. Stuttgart. E. Schweizerbart.

Thomas, L.

1931. Les Tumeurs des Poissons (Etude Anatomique et Pathogenique). *Assoc. Francaise pour L'Etude du Cancer.* 20: 703.

EXPLANATION OF THE PLATES.

Plate I.

Fig. 1. Osteochondroma on the right side o the head of a live aquarium-bre male jewelfish, *Hemichromis bimacu latus*. Enlarged about one-third. Pho tograph by S. C. Dunton, New Yor Zoological Society.

Fig. 2. Photomicrograph showing section o the osteochondroma taken throug the operculum. Note the thickene band of periosteal (**p**) material grow ing out from the surfaces of thi structure. The epithelium (**e**) varie in thickness but otherwise is norma in appearance. The corial (**c**) laye is edematous and shows no melano phores. The chondrocytes (**Cc**) shov an irregular pattern and the amoun of intercellular substance varies con siderably in density. Delafield's hema toxylin-eosin. About 75 ×.

Fig. 3. Section stained with Giemsa's. Not the varying density of intercellula material. **P**, Periosteum; **i**, intercellu lar material. About 75 ×.

Plate II.

Fig. 4. Section through the subperiostea region showing blood vessels an fibrous material. Note the granula debris within the larger blood vessel Mallory's Triple stain. About 160 ×

Fig. 5. A group of cells from the osteochon droma. Vacuole-like cytoplasmic struc tures are present similar to those shown in Text-figure 2. Giemsa's stain About 335 ×.

Fig. 6. Iron - hematoxylin treated section showing osteoid tissue staining black Smaller isogenic groups of cartilage cells may be seen at the periphery About 75×.

Plate III.

Fig. 7. Note the variability in size and shape in the structure of the chondrocytes. Giemsa's stain. 675 ×.

Fig. 8. Region of periosteum and subperiosteum. Transformation of the fibroblastic-like cells into chondrocytes is taking place, producing growth by apposition. Hematoxylin-eosin. 675 ×.

Plate IV.

Fig. 9. Connective tissue layer extending into the tumor forming the supporting stroma which provides the pathway for the blood vessels and other cellular elements. Giemsa's stain. 675 ×.

Fig. 10. Periphery of tumor showing collagenous fibers with spindle-shaped cells. Note transformation into chondrocytes. Giemsa's stain. About 675 ×.

Plate V.

Fig. 11. Osteoid tissue. Note the network of osteoblasts around the periphery of this tissue. Delafield's hematoxylin. About 675 ×.

Fig. 12. Same as Figure 11, showing details of osteoid formation. Delafield's hematoxylin. About 675 ×.

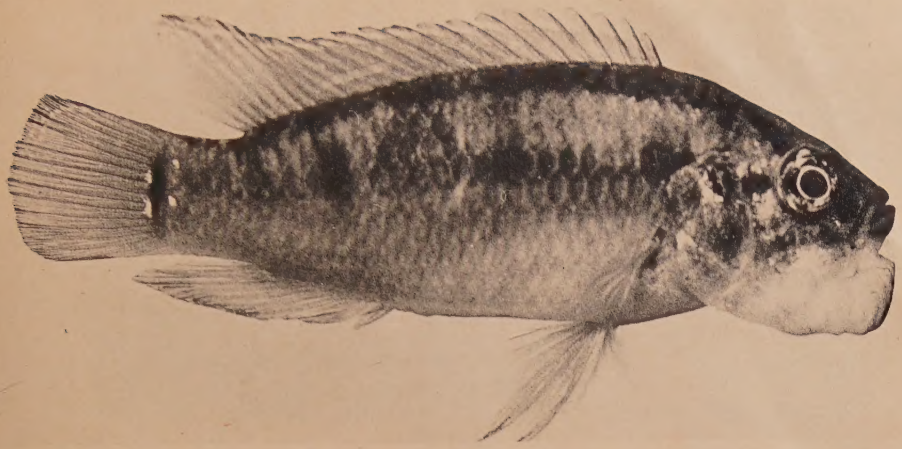


FIG. 1.

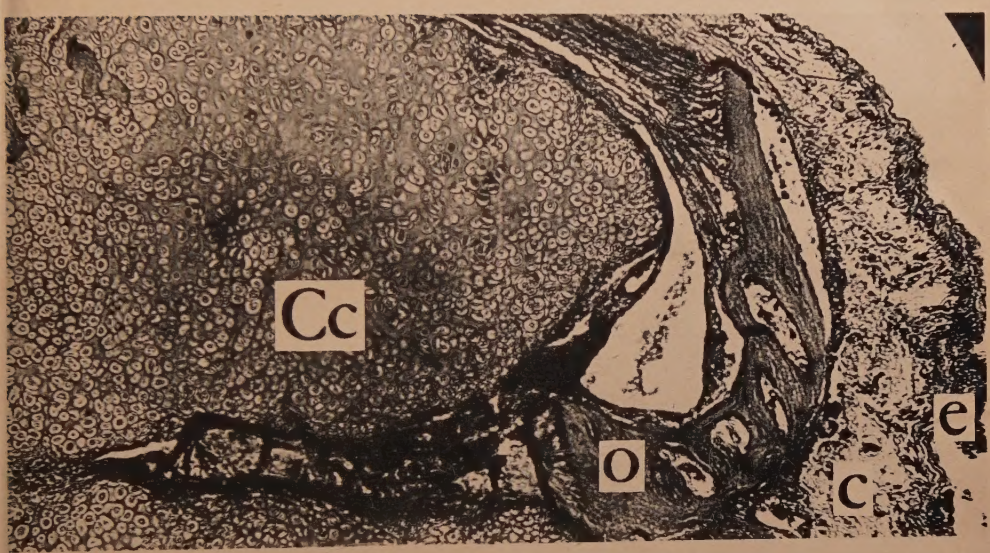


FIG. 2.

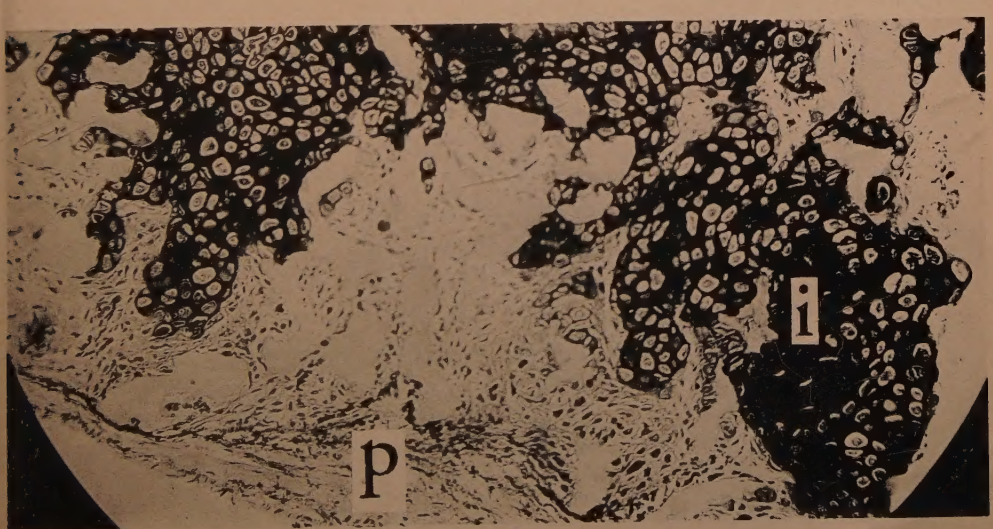
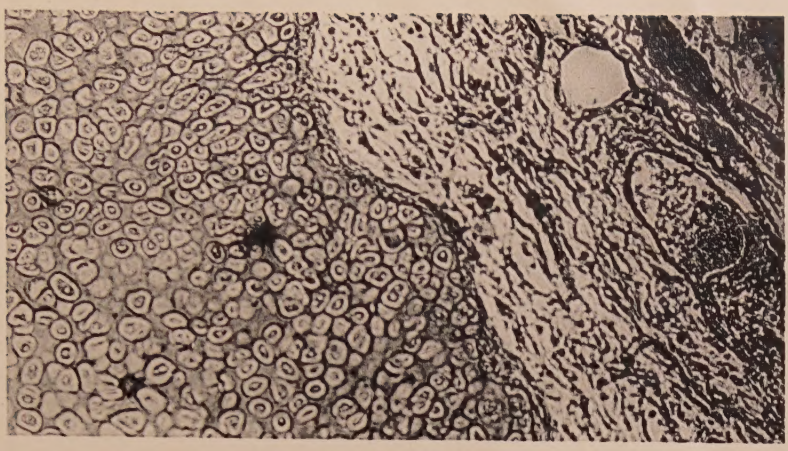


FIG. 3.

SPONTANEOUS NEOPLASMS IN FISHES. I. OSTEOCHONDROMA IN THE JEWELFISH, HEMICHROMIS BIMACULATUS.



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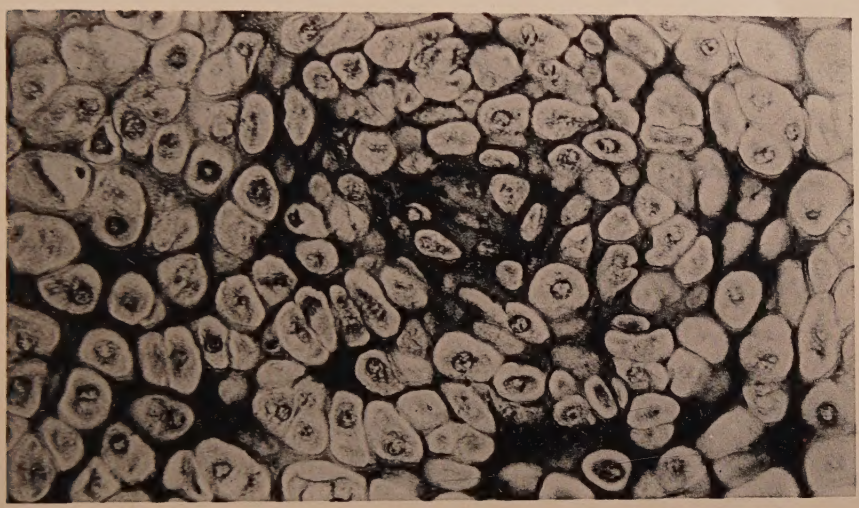

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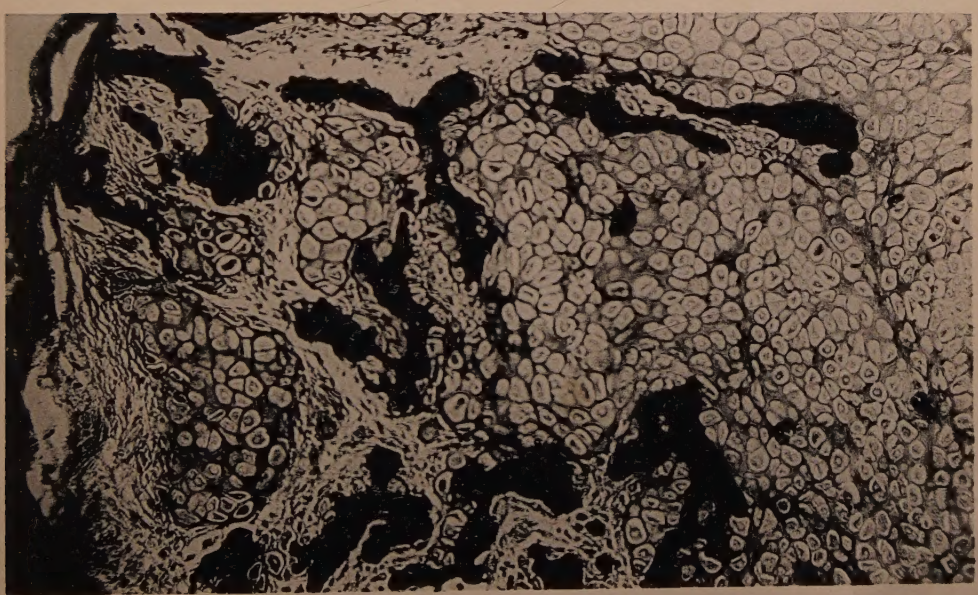

FIG. 6.

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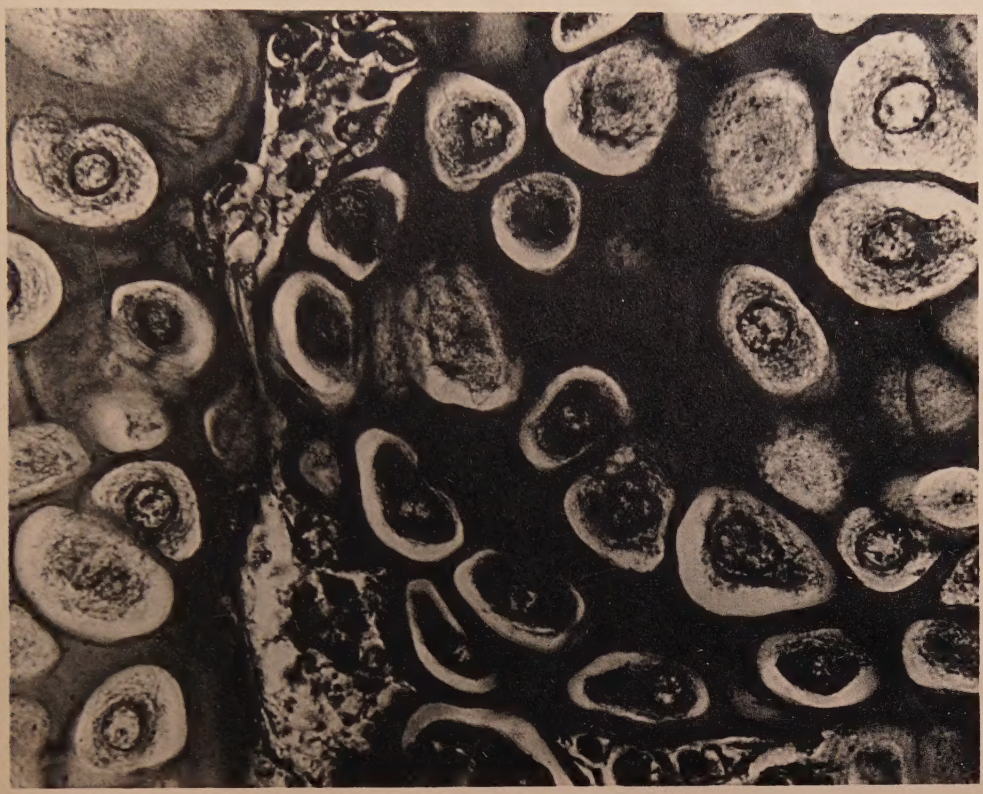

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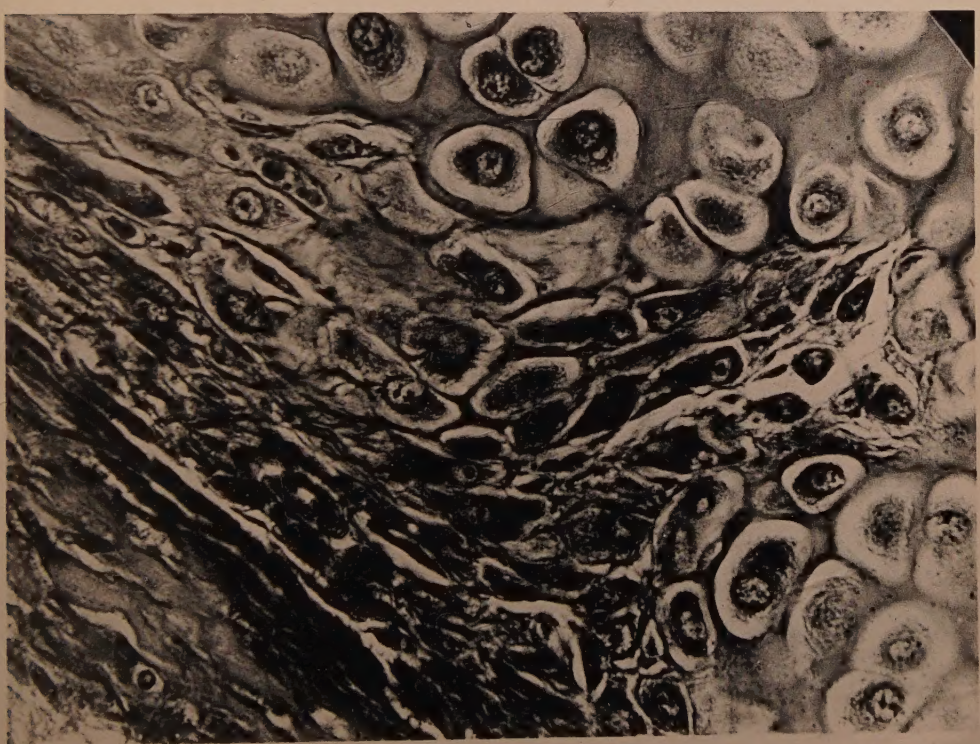

FIG. 8.

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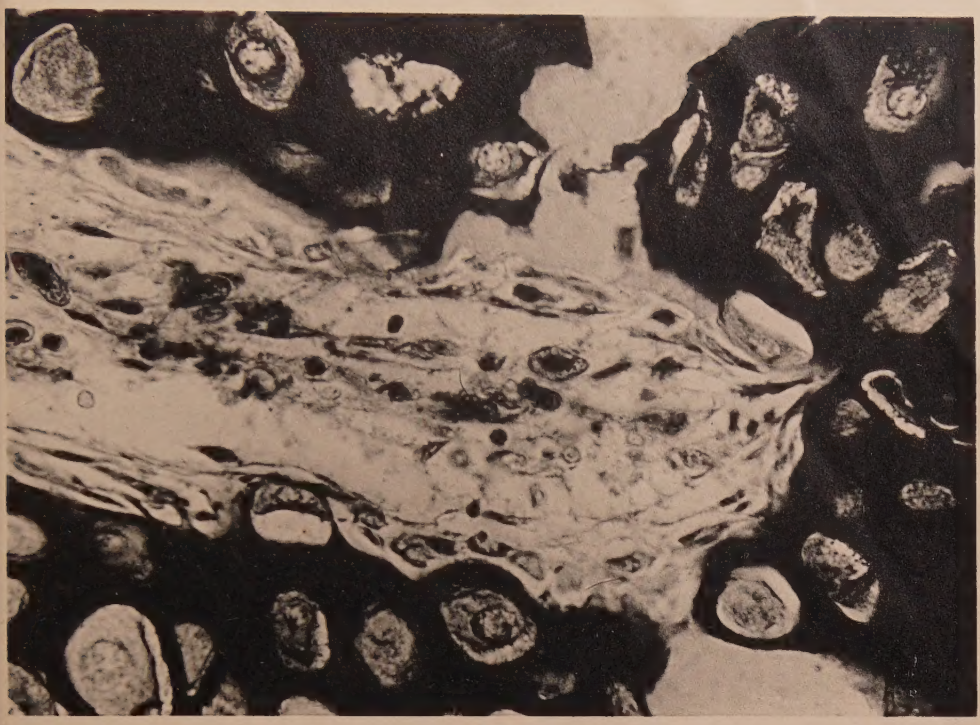

FIG. 9.

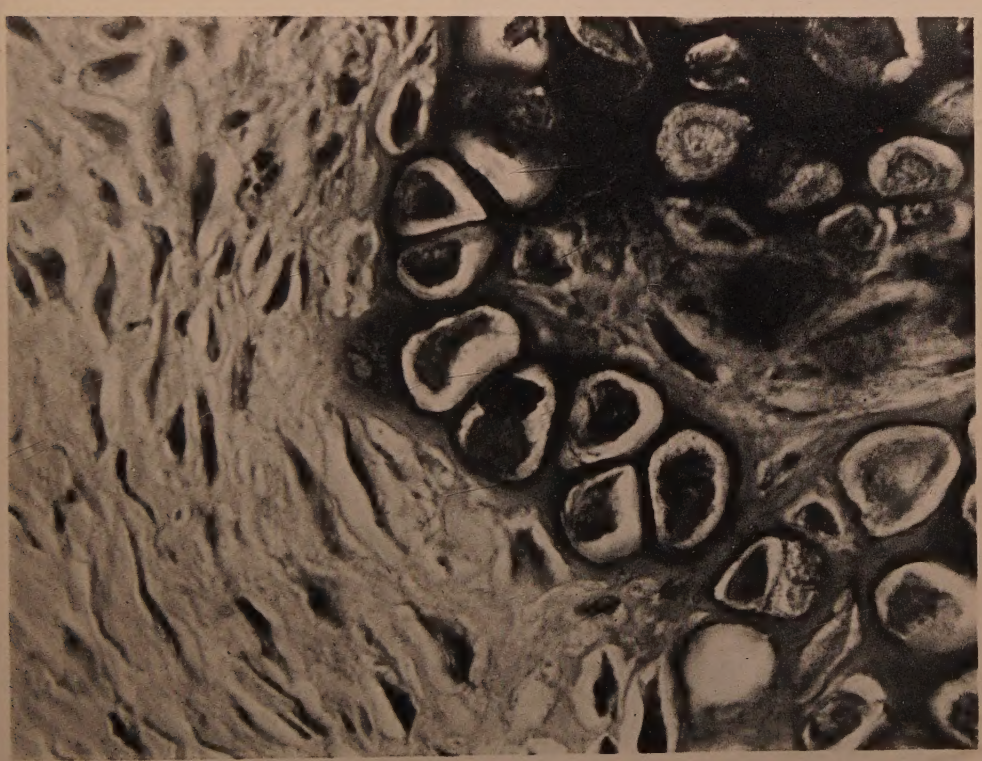

FIG. 10.

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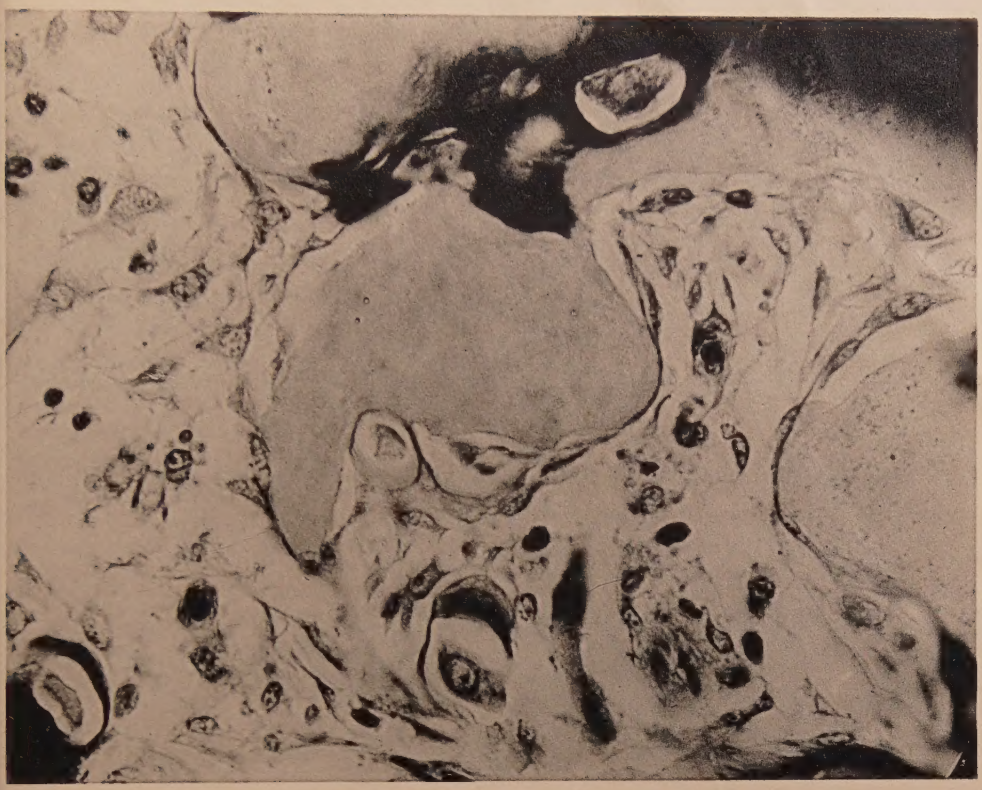

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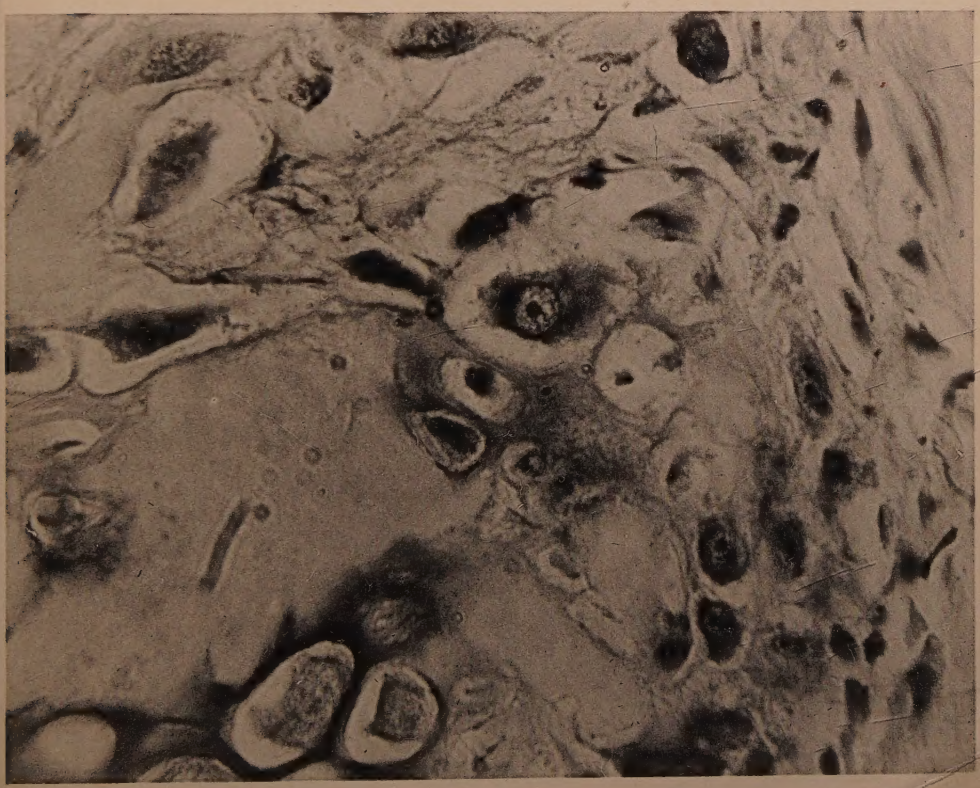

FIG. 12.

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